Introgression of improved fiber fineness trait in *G. hirsutum* L. from *G. longicalyx* Hutch. & Lee.

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Introduction

The cotton fiber quality has to be continually improved because of its large economic importance. Among all fiber properties, fineness is now considered with length and maturity as the main component of cotton fiber quality. According to N'Dungo et al., (1988) cotton fiber fineness could be improved by using the African wild diploid species *G. longicalyx* Hutch & Lee (2n=2x=26, F_1F_1). This study was conducted to confirm the karyotype of putative Monosomic Alien Addition Lines (MAALs) by cytogenetic and molecular analyses using GISH and SSR markers, and to evaluate the introgression of improved fineness in the backcross progenies of bi-specific and trispecific hybrids involving *G. longicalyx* through the measurement of the ribbon width of their fibers.

Materials and Methods

Plant material: The plants used in this study consisted in the progenies of MAALs and BC1/ BC2 plants developed according to the aphyletic(figure 1) and pseudophyletic (figure 2) methods described by Mergeai (2006). **Cytogenic identification**: The karyotype of presumed MAALs was etablished by chromosomes counting at mitotic metaphase and corroborated by GISH (Genomic *in situ* hybridization) according to D'hont et al (1995) protocols.

SSR markers analysis: 51 SSR reported by N'guyen et al (2004) were used to screen the plant material. **Ribbon Width measurement:** A sample of parallel fibers was cut and swelled in 18% NaOH (Figure 3) . The diameter of 200 fibers were then measured with an optical microscope and their ribbon width was determined by dividing the mean of the measured diameters by the 1.3 Summers coefficient (Roehrich, 1947).

Results and discussions

The counterstaining of the chromosome in plants mitotic cells revealed the presence of 52 + 1 extra chromosome in 9 lines on the ten analyzed (figure 2). The GISH performing on MAALs revealed 1 green chromosome corresponding to the alien chromosome of *G. longicalyx* (figure 4). Among the 51 polymorphic SSR used, 49 SSRwere present in the hexaploid (*G. hirsutum x G. longicalyx*)[2]. 18 SSR belonging to the linkage groups C1- C15, C4-C22, C6-25 and C10-C20 were found in the analyzed MAALs. Ribbon width measurement showed that *G. longicalyx* had the smallest ribbon width (6,41 µm) while the others genotypes showed ribbon width varying between 10 µm et 18 µm (Table 1).These results show that *G. longicalyx* contains genes that reduce the diameter of the fiber when associated to *G. hirsutum* genome.

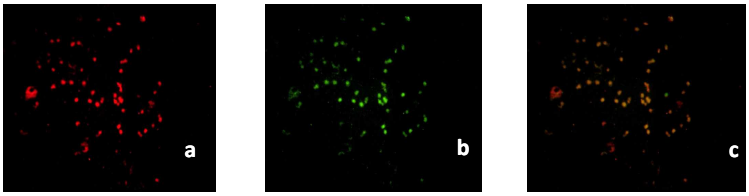

Figure 4 Genomic *in situ* hybridization on mitotic metaphase chromosomes.a)*The G. hirsutum* labelled-DNA linked with Texas-Red fluoresced red. b)*the G. longicalyx* coupled with FITC fluoresced green. c) The superposition of Texas Red and FITC detection.

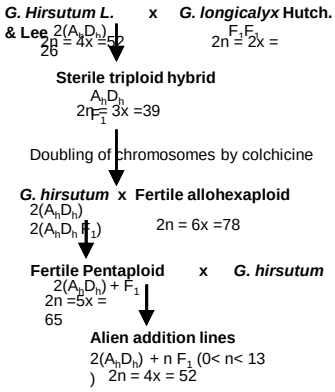

Figure 1. Scheme of the aphyletic method

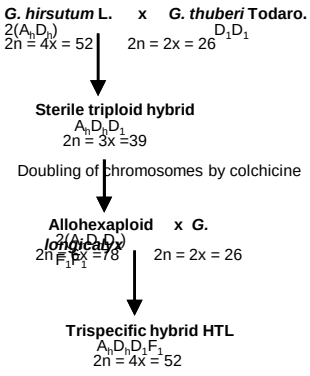

Figure 2. Scheme of the pseudophyletic method

Figure 3. Swelling of cotton fibers after treatment with 18% NaOH solution

Table 1. Ribbon width of analyzed plants

Genotype	Chr Nb	Genomic formula	Ribbon width in µm (standard deviation)
G.hirsutum var C2	52	$2(AD)_1$	17,45 (0,24)
G.hirsutum var NC8	52	$2(AD)_1$	15,60 (0,73)
G. longicalyx	26	$2F_1$	6,41 (0,42)
(G. hirsutum x G. longicalyx)[2]	78	$2(A_hD_hF_1)$	13,58 (0,40)
9052_10 LA 8 F1 I	53	$2(AD)_1 + 1$	14,86 (0,44)
9053_8 LA 9 F1 II	52	2(AD)1	14,07 (0,53)
9054_2 LA 10 F1 III	53	$2(AD)_1 + 1$	13,97 (0,60)
9055_3 LA 11 F1 IV	53	2(AD)1 + 1	16,89 (0,94)
9056_1 LA 12 F1 V	53	$2(AD)_1 + 1$	15,62 (0,61)
9057_3 LA 14 F1 VII	53	2(AD)1 + 1	15,54 (0,85)
9058_10 LA 15 F1 VIII	53	$2(AD)_1 + 1$	16,10 (1,42)
9059_2 LA 16 F1 IX	52	2(AD)1	13,85 (2,02)
9060_1 LA 18 F1 XI	53	$2(AD)_1 + 1$	13,09 (2,03)
9061_4 LA 19 F1 XII	53	$2(AD)_1 + 1$	15,17 (0,82)
9061_6 LA 19 F1 XII	52	2(AD)1	13,94 (0,80)
HTL3	52	AhDhD1F1	10,79 (2,44)
HTL - BC2_16 – BC2	52	AhDhD1F1	17,32 (0,18)
HTL - BC2_7 - BC2	**52**	**AhDhD1F1**	**11,98 (0,20)**
HTL - BC2_5 – BC2	52	AhDhD1F1	12,71 (0,04)
HTL BC1_6 – BC1	52	AhDhD1F1	12,33 (0,50)

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